Annex A: Water quality sampling and analysis plan, spring and summer 2016

Introduction

One of the benefits of a “Community Based Environmental Monitoring” (CBEM) project is to expand the monitoring capacity of government agencies and engage the public in environmental stewardship. (1) (2) In response to questions from the public and our own professional curiosity we have decided to initiate a limited CBEM project.

As part of the Montreux Clean Beach Project II(MCBPII), a water quality testing (WQT) program is being initiated at selected sites of the MCBPII. Symptoms reported by members and frequent bathers in the lake during previous summer seasons have sometimes been attributed to low bathing water quality, influenced by increased anthropogenic pressure of lakefront activities during certain times of the year. This year's WQT aims to gather microbiological data over a six week period in order to test this hypothesis.

Meetings at “La Maison de la Rivière” and the “Institut National de Recherche Agronomique” support the idea that the WQT information proposed is not regularly collected at the Haut-Lac locations covered by the MCBPII.

Objectives

CBEM programs are designed to educate the public, involve local citizens in environmental stewardship and provide reliable data for government and other interested organizations. In this regard the MCBPII CBEM program is no different than that of any other existing project. (3) (2) (4)

Specifically, the goals for this project are to:

1. Collect reliable water quality data as defined by the World Water Monitoring Challenge (5)
2. Collect and enumerate the quantity of key bacteriological indicators, in particular those related to fecal contamination (6)
3. Increase the skills of hammerdirt staff in regards to collecting and processing water samples.
4. Make the obtained data free and open to use for all concerned
5. Inform the public about affordable and simple water monitoring techniques
6. Inform the public of community based resources designed to advance scientific literacy

This project is a joint effort between hammerdirt association, biodesign.cc, AGiR!, and the Hackuarium.

Hackuarium

Action for Genomic integrity through Research

AGiR

Providing Information and Promoting Research

www.genomicintegrity.org

biodesign.cc

http://www.hackuarium.ch/fr/ http://www.hammerdirt.ch http://www.genomicintegrity.org/ https://biodesign.cc/

Deliverables

The objectives of a CBEM are designed to increase the monitoring capacity of current public systems and provide reliable, accessible data for the public and other government researchers. Data gathered from this project will be reported with our regular activity reports and the raw data will be available for download within 72 hours of the testing date from our repository on Github or through the KoboToolbox for the location in question. This document along with sampling protocols and all other supporting documents will be available on the wiki.

An end of project report will be published in PDF format, available for interested parties, and include:

1. Summary statistics and findings
2. Graphs/plots
3. Map overlay with densities and values
4. Comparison with government supplied data (if available)
5. Discussion of limitations and possible improvements to this CBEM

Testing materials

Turbidity, temp, DO and PH

Testing materials and supplies will be procured from the "World Water Monitoring Challenge"- http://www.monitorwater.org/ an international program managed by the "International Water Association" - http://www.iwa-network.org/ and the "Water Environment Federation" - http://www.wef.org/ .

Assembled and sold by Lamotte company as a "basic test kit," each kit includes one set of hardware and enough reagents to conduct up to 50 rounds of testing for pH, dissolved oxygen, temperature, and turbidity. Included are:

- 1 Instruction booklet (5)
- 1 Sample collection jar
- 1 pH test tube
- 1 Dissolved oxygen vial
- 1 Secchi disk decal
- 2 Temperature strips (14-40°C and 0-12°C)
- 50 pH reagent tablets (enough for 50 tests)
- 100 Dissolved oxygen reagent tablets (enough for 50 tests)
- 1 Color chart for determining DO, pH and turbidity test results
- 1 Mini pencil

Microbiological Analyses

Presence and quantity of *E.coli*, other coli forms, *Salmonella* and *Aeromonas* will be tested using "ECA Check Easygel". Easygel was first approved for "Water Watch" volunteer monitoring programs in the USA in 1999. A study released in 2009 compared the results of "Easygel" and "3M Petrifilm" used by volunteers to the results of laboratory analysis. Both methods had an overall accuracy rate above 80%. (7) (8) One big benefit of the Easygel format is that environmental samples of up to 5ml can be readily plated, with no worries about having to melt (and cool down!) agarose as used in standard microbial plates.

Members of the Hackuarium laboratory have used "Easygel" in previous educational activities, thus facilitating the training of hammedirt staff and volunteers.

Sampling methods

Water samples will be taken just below the surface (0.5m) and collected in sterile containers with a secure, air-tight lid.

Advantage: simple and inexpensive, an important consideration to a program on a budget. It also does not require any special sampling apparatus. The volunteer holds a sampling container under the surface of the water, expells visible air bubbles, and closes the container under the water (to avoids losing dissolved gasses, particularly important if testing for volatile analytes). (5)

Disadvantage: vertical gradients of temperature, oxygen, nutrients, and algae may be missed in these near surface samples. Deep lakes have a thermal gradient in the summer, and Lac Leman has at least two thermal regions (and, possibly, at least two chemical regions as well). Even in shallow, usually well-mixed lakes, chemical gradients will develop during calm periods.

Turbidity, temp, DO and PH

Two samples will be taken, one to be processed immediately and the other to be labeled and transported to Hackuarium for later analysis with the bioreporters from biodesign.cc

Microbiological samples

Three independent samples will be taken from each designated site in 15ml conical tubes pre-labeled in the following manner:

- Location
- Date
- Sample number for that day
- Time of sample

Once collected, the samples will be placed on ice and transported to the facility at Renens for plating on the ECA Check plates (http://www.micrologylabs.com/products.php?product_id=3) within 6 hours of the collection time. Label information will be recorded in the field notes and the “KoboToolBox” for that sampling date and location.

Processing samples and recording results

Turbidity, temp, DO and PH

The instructions provided with the kit (annex b) will be followed and the results will be noted in the field manual and entered into the “KoboToolbox” form for that sampling date and location.

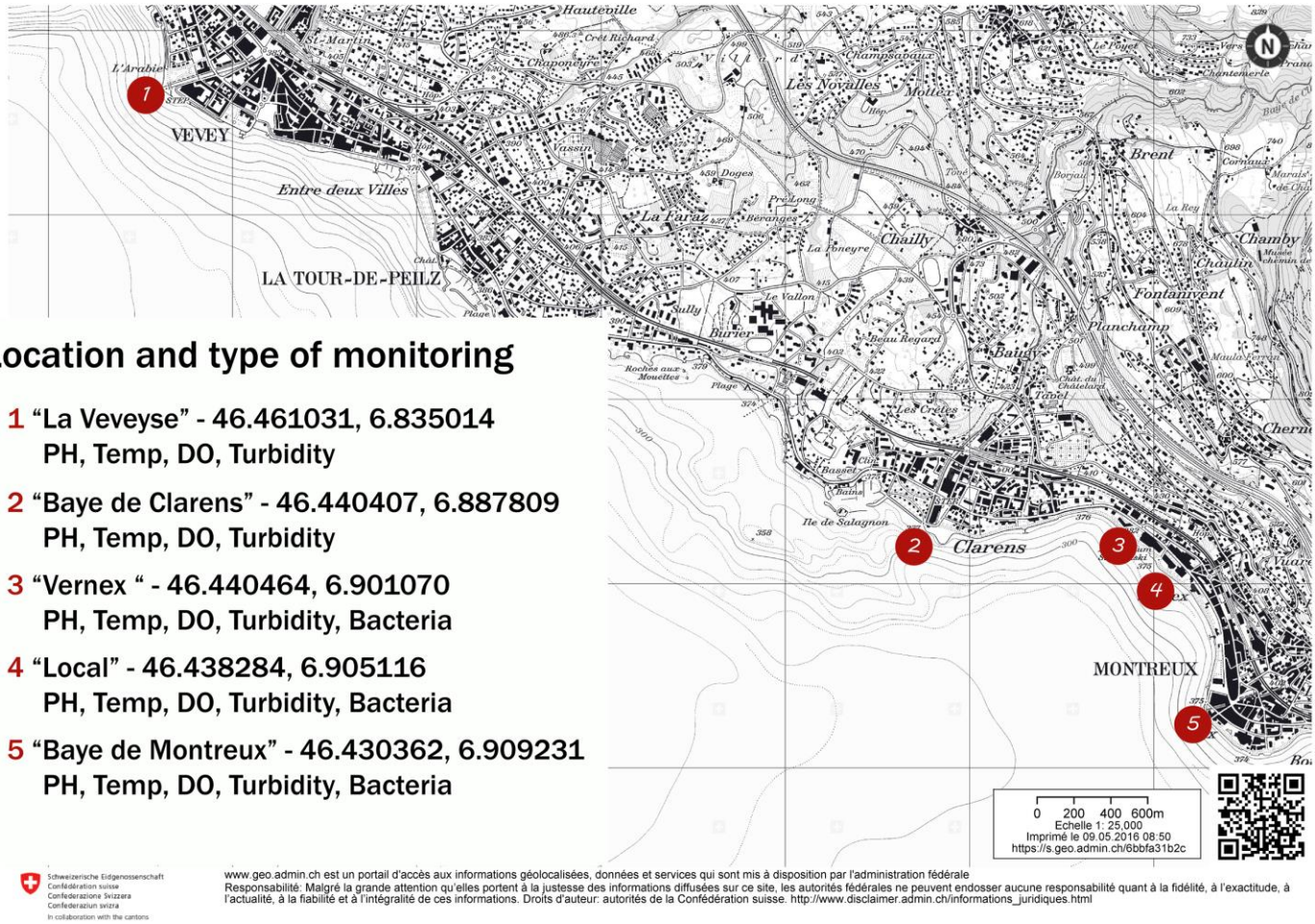
Microbiologicals

Petri dishes will be labeled in the same manner as the sample container with the addition of the quantity of sample added to the media. A 1-5ml of sample water will be mixed into the provided gel media solution, plated and incubated for about 18 hours at the Hackuarium facility in Renens.

After incubation, plates will be scored, counting total colony forming units (CFU), and each identifiable species (*E. coli* CFUs will appear dark blue, other coliform CFUs will appear lighter blue-gray or blue-violet, *Aeromonas spp.* will appear as pink to very light pink, and *Salmonella spp.* will appear as light green colonies). Data will be reported as CFU/100ml of sample.

With values from three independent samples at each site, means and standard errors can be used to test significance of the obtained data over the course of the sampling period.

Locations



Three of the five locations are currently part of the MCBPIIII beach-litter-survey project. Point “4” will be added to the beach litter survey by using a net to catch visible pollutants on the water surface below the boat dock. Point “3” will only be monitored for chemistry and biologicals. Correlations of the test results with the physical location of the sampling will be of great interest.

Testing Schedule

Microbiologicals

Testing will begin about 20 June 2016, and weekly samples will be taken from locations 3, 4 and 5. A 'negative control' sample will also be obtained each week from a local drinking fountain (labelled 'eau potable'). A positive control from an urban river (like the Vuachère or other) is also possible.

If a Tuesday afternoon testing protocol is followed over 7 weeks, the timeline would run from 21 June -2 August, require 21 sterile 15ml conical tubes for the water sampling, and 21 ECA Check EasyGel plates per site. After including a single 'negative control' for each sampling day, this will require 70 plates.

In case there are too many bacteria to count (TMTC) from certain samples, samples will be saved at 4^{o}C and further dilutions can be also plated and scored.

Turbidity, temp, DO and PH

Testing will begin on the Tuesday or Thursday the week the kits are received (late May). The kit contains enough reagents to conduct 50 tests, thus allowing us to monitor the locations for 10 weeks. Data will be carefully recorded, digitized and analyzed.

Field Blanks

Field blanks are used to assess potential contamination from sample handling, airborne materials, equipment, media, and other sources. A field blank usually consists of a sterile diluent sample of 2 mL that is taken to the site and poured into a properly labeled sample container during the first bacteria sampling event of that day. The blank sample is collected in the same type of container, labeled as a field blank, and handled and analyzed along with all the bacteria samples collected on that day. It is used to identify errors or contamination in sample collection and analysis.

Data

Handling and storage

The addition of water quality testing will require changes in our digital form and reporting methods. The form currently used by surveyors will change, thus separating the data in two sections, one section prior to WQT and one after. Data for the two sections will have to be merged prior to any analysis. The results of bacterial testing will be available 48-72 hours after sampling, thus adding another step to the survey. This should not, however, change the availability of the other survey results which is currently 24 hours after each survey.

Data analysis and presentation

The existing data set for MCBPII includes visible pollution surveys, hydrologic and meteorological data. With the addition of microbiological and basic water chemistry a more complete picture of the environmental status of the locations in question can be established.

Two interpretations of the microbiological data will be used: the geometric mean and the 95th percentile. The geometric mean is in common use in North America and the 95th percentile method conforms to EU directive 2006/7/EC. Furthermore, any publicly available water quality data in the area will be presented and integrated where appropriate.

The relationship between the results of WQT and the quantity and type of visible pollutants will be explored.

Partners and Responsibilities

PARTNER	ROLE
ACTION FOR GENOMIC INTEGRITY THROUGH RESEARCH!	Technical oversight for the microbial assays, training of hammerdirt staff, quality control of laboratory operations and of sample processing and analyses.
HACKUARIUM	Laboratory infrastructure and security, testing equipment and supplies for biologicals
HAMMERDIRT	Collection and processing of samples, data storage and administration. Communication of analyses and results.
BIODESIGN.CC	Analysis of samples for lead, mercury and arsenic using the bio-reporters

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